



**Image 1** Polymerase chain reaction amplification of various bacteria by acid-fast bacilli primers (**A**) or universal bacterial primers (**B**). Lane 1, 100-base-pair (bp) DNA ladder; lane 2, *Staphylococcus epidermidis*; lane 3, *Streptococcus pneumoniae*; lane 4, *Pseudomonas aeruginosa*; lane 5, *Neisseria lactamica*; lane 6, *Branhamella catarrhalis*; lane 7, *Escherichia coli*; lane 8, negative control without DNA template; lane 9, *Mycobacterium tuberculosis* (positive control). Note: This experiment was performed at the annealing temperature of 58°C (slightly more stringent) for 30 cycles, instead of the usual 55°C and 35 cycles. A very faint amplification band in lane 8 of Image 1B is seen, probably owing to the exquisite sensitivity of universal bacterial primers to amplify low number of copies of bacterial 16S ribosomal DNA in the buffer or reagent. This minute quantity of amplicon cannot be sequenced and does not interfere with other targeted amplification and subsequent sequencing.